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# Seven New *Eimeria* spp. (Protozoa, Coccidia) from Freshwater Fishes of Canada*

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SYNOPSIS. Seven new species of *Eimeria* are described and figured from the freshwater fishes of Ontario and Quebec, Canada. They are **Eimeria catostomi** sp. n. and **E. fernandoae** sp. n. from *Catostomus commersoni* (Lacépède), **E. etheostomae** sp. n. from *Etheostoma exile* (Girard), **E. hoffmani** sp. n. from *Umbra limi* (Kirtland), **E. micropteri** sp. n. from *Micropterus dolomieui* Lacépède **E. pungitii** sp. n. from *Pungitius pungitius* (Linnaeus), and **E. salvelini** sp. n. from *Salvelinus fontinalis* (Mitchill). Furthermore, 2 new host records and 2 new distribution records for North America are reported for *E. anguillae* Léger & Hollande, 1922 and *E. truttae* Léger & Hesse, 1919 respectively. Finally, morphologically similar oocysts found in various cyprinids are regarded as belonging to *E. iroquoina* Molnar & Fernando, 1974.

**Index Key Words:** *Eimeria catostomi* sp. n.; *Eimeria etheostomae* sp. n.; *Eimeria fernandoae* sp. n.; *Eimeria hoffmani* sp. n.; *Eimeria micropteri* sp. n.; *Eimeria pungitii* sp. n.; *Eimeria salvelini* sp. n.; *Eimeria anguillae*; *Eimeria truttae*; *Eimeria iroquoina*; Canadian freshwater fishes; light microscopy; taxonomy.

IN a previous article 12 new species of *Eimeria* were described from the freshwater fishes of Ontario. A further investigation of fishes in Ontario and Quebec has yielded additional new species of *Eimeria*.

Seven are described and figured and new host records for species that were previously found are reported.

## MATERIALS AND METHODS

The host specimens were collected during the summer of 1973 from Grand River system, Ontario (Grand River, Conestogo River, Laurel Creek); from Bronte Creek, Ontario; from Saugeen River near Durham, Ontario; and from Matamek River, Quebec. The fish were transported to the laboratory alive and examined immediately, or they were kept for several days in the holding tanks, allowing the content of the gut to pass. Squash preparations of the different inner organs and the mucus and scrapings of the epithelial wall of the intestine were made. The material, on microslides with coverslip, was examined with a Leitz Orthoplan microscope under oil immersion. At least 30 oocysts of each species were measured and drawings made. Positive slides were fixed in 2.5% (v/v) glutaraldehyde solution by irrigation. The coverslips were sealed with Canada balsam to prevent desiccation. Under these conditions the parasites retained their original shape for a period of a few weeks during which time they could be studied in detail. **All measurements are in micrometers.**

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## RESULTS AND DISCUSSION

### **Eimeria catostomi** sp. n.

(Figs. 1, 1a)

*Diagnosis.*—Oocyst spherical, 7.0 (6.5-7.5) in diameter. Cyst wall smooth, colorless, composed of a single, very thin layer (~0.1). Sporocysts compact, in contact with the oocyst wall. Oocyst residuum and micropyle absent, polar granule in the center of the oocyst, measuring 0.8 (0.7-0.9). Sporocysts oval, flattened from one side, and measuring 5.5 (5.2-5.9) in length, 3.8 (3.6-4.0) in width, and 3.1 (2.9-3.4) in depth. Sporocyst wall single layered, 0.2 thick; Stieda body absent. Sporocysts arranged 3 in 1 plane, 4th overlying. Two vermiform crescent-shaped sporozoites with one end reflected, arranged head to tail in the sporocyst, measuring 4.6 (4.4-4.8) × 1.4 (1.3-1.6). Refractile globule in sporozoites absent. Sporocyst residuum is round, finely granulated, and measuring 2.3 (2.0-2.6) in diameter.

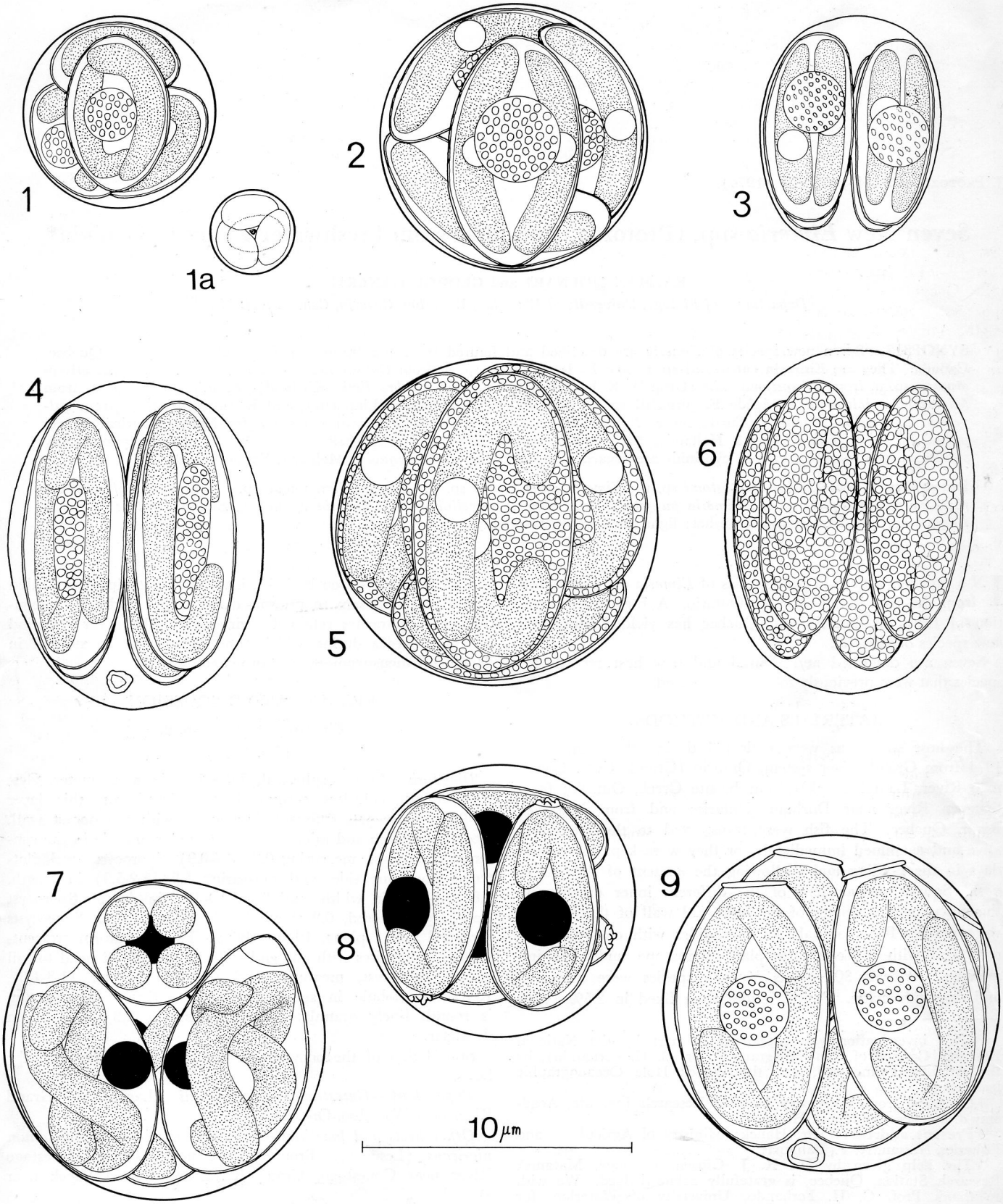
Sporulation of the oocysts was completed in the gut of the hosts.

*Type host.*—*Catostomus commersoni* (Lacépède). Grand River near Waterloo, Ontario.

*Other hosts and localities.*—*C. commersoni* and *Hypentelium nigricans* (Lesueur). Bronte Creek near Milton, Conestogo River near Conestogo, Grand River and Laurel Creek near Waterloo; all in Ontario.

*Location in host.*—Epithelium of gut's anterior portion, feces.

*Host infection ratios.*—14/9, *C. commersoni*; 4/2, *H. nigricans.*




Figs. **1-9**. *Eimeria catostomi* sp. n. 1a. Position of the polar granule in the oocyst. **2**. *Eimeria etheostomae* sp. n. **3**. *Eimeria fernandoae* sp. n. **4**. *Eimeria hoffmani* sp. n. **5**. *Eimeria micropteri* sp. n. **6**. *Eimeria pungitii* sp. n. **7**. *Eimeria salvelini* sp. n. **8**. *Eimeria anguillae* Léger & Hollande, 1922. **9**. *Eimeria truttae* Léger & Hesse, 1919.

*Remarks.*—Oocysts of *E. catostomi* fall into the size range of *E. haneki* Molnar & Fernando, 1974, but differ from them by the arrangement of the sporocysts and polar granules. The sporocysts of *E. haneki* are usually arranged in 2 and 2 combination and 1 or 2 polar granules are located on the periphery in the oocyst. The sporocysts of *E. catostomi*, however, are always arranged in 3 and 1 combination and a single polar granule is located in the center of the oocyst as shown in Fig. 1a.

**Eimeria etheostomae** sp. n.

(Fig. 2)

*Diagnosis.*—Oocyst spherical, 9.4 (9.1-10.4) in diameter. Cyst wall smooth, colorless, composed of a single, very thin layer (~0.1). Sporocysts compact, in contact with the oocyst wall. Oocyst residuum, micropyle and polar granule absent. Sporocysts coffee-bean-shaped, measuring 8.5 (7.8-9.1) in length, 5.0 (4.5-5.4) in width, and 4.7 (4.2-5.0) in depth; Stieda body absent. Sporocysts usually arranged 3 in 1 plane, 4th overlying. Two banana-shaped sporozoites arranged head to tail in the sporocyst, measuring 8.4 (7.8-9.0) × 1.6 (1.4-1.8). Rarely, one end of the sporozoite may be reflexed; i.e. bent over at the tip. Each sporozoite possesses a round refractile globule. Sporocyst residuum finely granulated, round, 3.6 (3.1-3.9) in diameter.

About half the oocysts were sporulated in the host. Others were passed in the semisporulated condition.

*Type host.—Etheostoma exile* (Girard). Laurel Creek near Waterloo, Ontario.

*Other hosts and localities.—E. exile* and *E. nigrum* Rafinesque. Bronte Creek near Milton and Laurel Creek near Waterloo, all in Ontario.

*Location in host.*—Epithelium of gut's anterior portion, feces.

*Host/infection ratios.*—8/10, *E. exile*; 7/4, *E. nigrum.*

**Eimeria fernandoae** sp. n.

(Fig. 3)

*Diagnosis.*—Oocyst ellipsoid, 8.3 (7.8-9.0) long and 6.6 (6.5-7.0) wide. Cyst wall smooth, colorless, composed of a single, very thin layer (~0.1). Sporocysts compact, in contact with the oocyst wall. Oocyst residuum, micropyle and polar granule absent. Sporocysts elongatedly ellipsoid, measuring 7.2 (6.8-7.5) × 3.0 (2.6-3.4). Sporocyst wall 0.2 thick; Stieda body absent. Four sporocysts arranged lengthwise and in the same direction in the oocyst. Each sporocyst with 2 banana-shaped sporozoites arranged head to tail and measuring 5.8 (5.2-6.5) × 1.2 (1.0-1.3). Each sporozoite possesses a round refractile globule. Sporocyst residuum round, finely granulated, and 2.6 (2.3-3.2) in diameter.

Sporulation of the oocysts was completed in the gut of the hosts.

*Type host.—Catostomus commersoni* (Lacépède). Grand River near Waterloo, Ontario.

*Other hosts and localities.—C. commersoni* and *Hypentelium nigricans* (Lesueur). Bronte Creek near Milton, Conestogo River near Conestogo, Grand River near Waterloo; all in Ontario.

*Location in host.*—Epithelium of gut's anterior portion, feces.

*Host/infection ratios.*—14/8, *C. commersoni*; 4/3, *H. nigricans.*

*Remarks.*—This species is named in honor of Dr. M. A. Fernando, Ontario Veterinary College, University of Guelph, Guelph, Ontario.

**Eimeria hoffmani** sp. n.

(Fig. 4)

*Diagnosis.*—Oocyst ellipsoid, 11.5 (11.0-12.2) long and 9.2 (9.1-9.6) wide. Cyst wall smooth, colorless, composed of a single, very thin layer (~0.1). Sporocysts moderately compact. Oocyst residuum and micropyle absent, one polar granule present, measuring 0.8 (0.7-0.9). Sporocysts elongatedly ellipsoid, measuring 10.0 (9.6-10.4) × 3.6 (3.4-3.9). Sporocyst wall 0.2 thick; Stieda body absent. Four sporocysts arranged lengthwise and in the same direction in the oocyst. Each sporocyst with 2 vermiform sporozoites arranged head to tail. One end of all sporozoites reflexed. Measurements of sporozoites (without reflexed portion) 8.6 (8.4-8.7) × 1.4 (1.3-1.5). Refractile globule in sporozoites absent. In the young oocysts, sporocyst residuum elongatedly ellipsoid, finely granulated, and measuring 6.4 (6.2-6.6) × 2.0 (1.8-2.2). In the older oocysts the residuum is either less elongated or appears scattered.

Sporulation of the oocysts was completed in the gut of the host.

*Type host.—Umbra limi* (Kirtland). Bronte Creek near Milton, Ontario.

*Location in host.*—Epithelium of gut's anterior portion, feces.

*Host/infection ratio.*—8/8.

*Remarks.—Eimeria hoffmani* differs from *E. fernandoae* by its larger oocysts, moderately compact sporocysts, reflexed sporozoites, and by possessing a polar granule. This species is named in honor of Dr. G. L. Hoffman, Eastern Fish Disease Laboratory, Kearneysville, W. Va., U.S.A.

**Eimeria micropteri** sp. n.

(Fig. 5)

*Diagnosis.*—Oocyst spherical, 12.0 (11.7-12.5) in diameter. Cyst wall smooth, colorless, composed of a single, very thin layer (~0.1). Sporocysts compact, in contact with the oocyst wall. Oocyst residuum, micropyle and polar granule absent. Sporocysts ellipsoid, measuring 11.4 (11.0-11.7) × 6.2 (6.0-6.5). Sporocyst wall 0.2 thick; Stieda body absent. Sporocysts arranged 3 in 1 plane, 4th overlying. Each sporocyst with 2 vermiform sporozoites arranged head to tail. One end of all sporozoites reflexed. Measurements of sporozoites (without reflexed portion) 9.1 (8.9-9.3) × 2.1 (2.0-2.2). Each sporozoite possesses a round refractile globule. Sporocyst residuum finely granulated, dispersed.

Most of the oocysts (~90%) leave the gut semisporulated, the rest being fully sporulated.

*Type host.—Micropterus dolomieui* Lacépède. Conestogo River near Conestogo, Ontario.

*Other hosts and localities.—M. dolomieui* and *M. salmoides* (Lacépède). Conestogo River near Conestogo and Laurel Creek near Waterloo; all in Ontario.

*Location in host.*—Epithelium of gut's anterior portion, feces.

*Host/infection ratios.*—7/3, *M. dolomieui*; 17/8, *M. salmoides.*

*Remarks.*—This species differs from *E. laureleus* Molnar & Fernando, 1974 and *E. ojibwana* Molnar & Fernando, 1974 by its elliptical sporocysts.

**Eimeria pungitii** sp. n.

(Fig. 6)

*Diagnosis.*—Oocyst ellipsoid, 12.5 (12.1-13.0) long and 9.8 (8.6-10.4) wide. Cyst wall smooth, colorless, composed of

a single, very thin layer (~0.1). Sporocysts compact, in contact with the oocyst wall. Oocyst residuum, micropyle and polar granule absent. Sporocysts elongatedly ellipsoid, measuring 10.0 (9.1-10.4) × 3.6 (3.4-3.9). Sporocyst wall 0.2 thick; Stieda body absent. Four sporocysts arranged lengthwise and in the same direction in the oocyst. Each sporocyst with 2 banana-shaped sporozoites arranged head to tail in the sporocyst, measuring 8.7 (8.4-9.1) × 2.3 (2.1-2.4). Each sporozoite possesses a round refractile globule. Sporocyst residuum granulated, dispersed, and completely fills the sporocyst.

Only a small portion of the oocysts complete sporulation in the host's gut and most of them leave the fish unsporulated. The description, however, was based on those oocysts found fully sporulated.

*Type host.—Pungitius pungitius* (Linnaeus). Matamek River, Quebec.

*Location in host.*—Epithelium of gut's anterior portion, feces.

*Host/infection ratio.*—33/17.

*Remarks.—E. pungitii* differs from *E. fernandoae* and *E. umbrae* by its sporulation habit. It differs furthermore, from *E. umbrae* by its banana-shaped sporozoites and the absence of the polar granule, from *E. fernandoae* by possessing larger oocysts.

**Eimeria salvelini** sp. n.

(Fig. 7)

*Diagnosis.*—Oocyst spherical, 12.0 (11.7-12.5) in diameter. Cyst wall smooth, colorless, composed of a single, very thin layer (~0.1). Sporocysts compact, in contact with the oocyst wall. Oocyst residuum, micropyle and polar granule absent. Sporocysts oval, measuring 9.2 (9.0-9.4) × 5.1 (5.0-5.3). Cap-like Stieda body at its tapered end 1.1 (0.9-1.3) long. Sporocyst wall 0.2 thick. Sporocysts arranged 3 in 1 plane, 4th overlying. Each sporocyst with 2 vermiform sporozoites measuring 6.8 (6.5-7.1) × 1.5 (1.3-1.7), which are characteristically interlaced. Refractile globule in sporozoites absent. Sporocyst residuum round, compact, and measuring 1.8 (1.5-2.1) in diameter.

Sporulation of the oocysts was completed in the gut of the host.

*Type host.—Salvelinus fontinalis* (Mitchill). Matamek River, Quebec.

*Location in host.*—Epithelium of gut's anterior portion, feces.

*Host/infection ratio.*—10/2.

*Remarks.*—This species resembles *E. tedlai* Molnar & Fernando, 1974, but differs from it in the larger size of the oocysts, by not possessing a polar granule, and by the shape of the sporozoites.

*Eimeria anguillae* Léger & Hollande, 1922

(Fig. 8)

*Diagnosis.*—Oocyst spherical, 9.8 (9.1-10.4) in diameter. Cyst wall smooth, colorless, composed of a single, very thin layer (~0.1). Sporocysts compact, in contact with the oocyst wall. Oocyst residuum micropyle and polar granule absent. Sporocysts oval, measuring 7.8 (7.5-8.0) × 4.1 (3.9-4.4). Stieda body formed by the 4 papillae situated on the tapered end of the sporocyst. Sporocyst wall 0.2 thick. Sporocysts arranged in 2 parallel pairs crossing each other. Each sporocyst with 2 vermiform sporozoites arranged head to tail; one end of each reflexed. Measurements of sporozoites (without reflexed portion) 6.8 (6.5-7.0) × 1.5 (1.3-1.8). Refractile globule in sporozoites absent. Sporocyst residuum round or ellipsoid, compact, and measuring 2.3 (1.9-2.6) × 1.6 (1.3-1.9).

Sporulation of the oocysts has been completed in the gut of the host.

*Host.—Anguilla rostrata* (Lesueur). Matamek River, Quebec.

*Location in host.*—Epithelium of gut's anterior portion, feces.

*Host/infection ratio.*—33/2.

*Remarks.—Eimeria anguillae* is known from *A. anguilla* Linnaeus in Europe. The present discovery constitutes a new host record and extends its known distribution to North America.

*Eimeria truttae* Léger & Hesse, 1919

(Fig. 9)

*Diagnosis.*—Oocyst spherical, 12.8 (12.3-13.0) in diameter. Cyst wall smooth, colorless, composed of a single, very thin layer (~0.1). Sporocysts compact, in contact with the oocyst wall. Oocyst residuum and micropyle absent, a single polar granule present, 0.9 (0.7-1.0) in diameter. Sporocysts oval, measuring 10.1 (9.3-11.0) × 6.0 (5.0-6.5). Disc-like Stieda body 2.7 (2.6-2.8) in diameter at its tapered end. Four sporocysts arranged in the same direction in the oocyst. Each sporocyst with 2 vermiform sporozoites arranged head to tail; one end of each reflexed. Measurements of sporozoites (without reflexed portion) 8.5 (7.8-9.0) × 1.6 (1.3-2.0). Refractile globule in sporozoites absent. Sporocyst residuum finely granulated, round, and measuring 2.8 (2.6-3.0) in diameter.

Sporulation of the oocysts was completed in the gut of the host.

*Host.—Salvelinus fontinalis* (Mitchill). Matamek River, Quebec.

*Location in host.*—Epithelium of gut's anterior portion, feces.

*Host/infection ratio.*—10/2.

*Remarks.*—This parasite is known from *Salmo trutta fario* Linnaeus in Europe. The present discovery is a new host record and extends its known distribution to North America.

*Eimeria iroquoina* Molnar & Fernando, 1974

*Hosts and infection ratios.—Nocomis biguttatus* (Kirtland), 17/2; *Notropis heterolepis* Eigenmann & Eigenmann, 17/2; *N. rubellus* (Agassiz), 7/6; *Pimephales notatus* (Refinesque), 18/8; *Rhinichthys atratulus* (Hermann), 6/4; *Semotilus atromaculatus* (Mitchill), 20/2.

*Localities.*—Bronte Creek near Milton, Conestogo River near Conestogo, Grand River and Laurel Creek near Waterloo, Sauge River near Durham; all in Ontario.

*Location in hosts.*—Epithelium of gut's anterior portion, feces.

*Remarks.*—The oocysts found in the guts of the above Cyprinids fall into the size range of *E. iroquoina* described from *Notropis cornutus* (Mitchill), but differ from the type species in some respects, e.g. by the shape of sporocysts and sporocyst residuum, and by the presence or absence of either polar granules in the oocysts or of refractile globules in the sporozoites (Table 1). Because of the close relationship of the hosts and mainly because of almost indistinguishable differences in the oocysts structure, however, these oocysts must be regarded as belonging to *E. iroquoina* until further intensive examinations are made.

The great majority of the species of *Eimeria* that are so

Table 1. *Variation of* Eimeria iroquoina *Molnar & Fernando, 1974, in various cyprinid hosts.*

| Morphological Criteria | *Notropis cornutus* | *Notropis heterolepis* | *Notropis rubellus* | *Semotilus atromaculatus* | *Rhinichthys atratulus* | *Nocomis biguttatus* | *Pimephales notatus* |
|---|---|---|---|---|---|---|---|
| Oocyst size* | 9.0-11.0 | 9.4-10.4 | 8.7-10.4 | 9.1-9.8 | 9.0-9.5 | 10.4-10.8 | 8.6-1.0 |
| Polar granule† | − | + | − | − | + | − | − |
| Length of Sporocysts* | 7.8-8.0 | 6.8-7.2 | 7.3-8.0 | 7.2-7.8 | 7.6-8.0 | 6.5-7.0 | 8.6-10.0 |
| Width of Sporocysts* | 4.0-4.5 | 4.1-4.4 | 4.0-5.0 | 4.0-5.0 | 4.5-4.9 | 3.9-4.9 | 3.3-4.0 |
| Sporocysts shape | ellipsoid | bluntly ellipsoid | ellipsoid | tapered ellipsoid | tapered ellipsoid | bluntly ellipsoid | bluntly ellipsoid |
| Arrangement of sporocysts | 2-2 | 2-2 or 3-1 | 2-2 or 3.1 | 3-1 | 3-1 | 3-1 or 2.2 | 2-2 |
| Length of sporozoites* | 7.5-7.7 | 5.0-5.3 | 5.5-6.5 | 5.0-5.6 | 6.5-7.2 | 4.7-5.2 | 4.5-5.2 |
| Width of Sporozoites | 1.5-1.7 | 1.3-1.5 | 1.3-2.2 | 1.3-1.5 | 1.3-1.6 | 1.3-1.6 | 1.3-1.6 |
| Refractile globule† | + | + | − | − | + | − | + |
| Sporocyst residuum shape | round or ellipsoid | ellipsoid | round or ellipsoid | round | round | round | oval |
| Residuum granulation | fine | fine | fine | fine | fine | fine | fine |
| Residuum size* | 2.3-4.0 by 2.2-3.0 | 3.2-4.3 by 2.0-2.6 by 1.5-2.0 | 2.0-3.2 by 2.0-2.2 | 1.7-2.2 | 1.9-2.2 | 1.8-2.6 | 2.6-2.9 by 1.8-2.1 |

* Measurements in micrometers. † +, present; −, absent.

far known from fish, complete their sporulation in the gut of their hosts (4, 5), the only known exceptions being *E. pigra* (2) and *E. aurati* (1). The situation in *E. etheostomae*, *E. micropteri*, and *E. pungitii* indicates that the number of Eimerias which complete their sporulation outside the host may be greater than has so far been thought. Unfortunately, an attempt to cause sporulation by placing unsporulated oocysts into 2.0 or 2.5% (w/v) $K_2Cr_2O_7$ solution failed. In contrast to the coccidia of mammals and birds, oocysts from fish are very sensitive to changes in the concentration of surrounding solutions (salt, glycerol, sugar) and rapidly lose their shape and die.

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